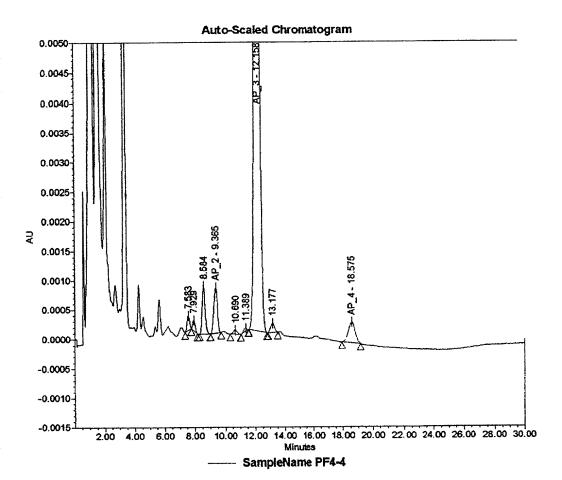


Ansamitocin P-3 (mg/L)

Lyophile (or frozen) culture Place on agar plate with medium CM4-1 28 °C, 5-7 days Pinpoint transfer to another CM4-1 agar plate 28 °C, 7 days Macerate each one colony in a screwed tube containing 10 glass bead (2mm dia.) and 2 mL H₂O Inoculate each 0.5 mL colony suspension to the seed (medium fill volume: 30 mL/250 mL flask) 28 °C, 48 hours shaker (throw: 70 mm; rotary speed: 220 rpm) Transfer 1.0 mL grown seed to fermentation medium (fill volume: 20 mL/250 mL flask) 28 °C, 8 days shaker (throw: 70 mm; rotary speed: 220 rpm) Harvest Use 4.5 mL ethanol (95%) to extract each 0.5 mL whole broth Mix (extract for 10 min on a vortex mixer (SMI model 2600) Centrifuge Supernatant for hplc assay



G4 AP-3

		Component	04_78*0			
	Sample		RT	Area	Height	Amount
1	PF4-4		7.583	2809	251	
2	PF4-4		7.929	2168	178	
3	PF4-4		8.584	11415	818	
4	PF4-4	AP_2	9.365	12486	785	14.062
5	PF4-4		10.690	1354	71	
6	PF4-4		11.389	821	56	
7	PF4-4	AP_3	12.158	345473	17000	389.092
8	PF4-4		13.177	3001	160	
9	PF4-4	AP_4	18.575	10938	357	12.319